

## 41. The Preparation of 3:5-Dinitrobenzoic Acid and 3:5-Dinitrobenzoyl Chloride

Observations on the Acylation of Amino-acids by means of 3:5-Dinitrobenzoyl Chloride and certain other Acid Chlorides

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3:5-Dinitrobenzoyl chloride was suggested by one of us [Saunders, 1934; 1938] as a useful reagent for the identification of certain classes of amino-acids. The preparation of this reagent has now been examined in detail. The two recorded preparations of 3:5-dinitrobenzoic acid from benzoic acid [Shukow, 1895; Sirks, 1908] are long, the former author recommending the heating of benzoic acid with fuming nitric acid and concentrated sulphuric acid for 8–10 hr. on a water bath and then for 1–2 hr. on a sand bath. The second author heated the mixture on a water bath for 12 hr., poured into water and then repeated the nitration on the product for a further 12 hr. Details are given below for the preparation of 3:5-dinitrobenzoic acid in which, by careful temperature control, the period of nitration for a corresponding weight of benzoic acid has been reduced to  $2\frac{1}{2}$  hr. Moreover, Sirks's yield has been improved upon and the quantities of nitrating acid used by Shukow have been reduced by 15%.

The recorded preparation of 3:5-dinitrobenzoyl chloride [Berend & Heymann, 1904] consists of heating the acid with phosphorus pentachloride and ultimately distilling the product under reduced pressure at the rather high temperature of 200°. During this process, the 3:5-dinitrobenzoyl chloride is liable to explode. Furthermore, the separation of the solid acid chloride in the condenser may be very troublesome. A more convenient method is now described in which the final distillation is avoided. The pure product has m.p. 69.5° which is close to that of 68–69° given by Eastman Kodak [1934]. Cohen & Armes [1906] give 66–68°. The higher m.p. of 74° obtained by Berend & Heymann [1904] and by Town [1941] has not been observed by us, even after distillation at 172° and 15 mm.

The method of 3:5-dinitrobenzoylation used in the above communications [Saunders, 1934; 1938] was rapid and simple and consisted of dissolving the amino-acid in *N* NaOH and shaking the solution for about 2 min. with finely powdered 3:5-dinitrobenzoyl chloride. The latter dissolved extremely rapidly, and on acidification the 3:5-dinitrobenzoyl derivative was precipitated.

It was found that neutral and basic ampholytes gave crystalline derivatives in good yield, whereas acidic ampholytes were usually less reactive, and, under the conditions of the experiment, glycine could be separated, for example, from aspartic acid. Tyrosine gave no derivative.

It was also shown that the reagent could be used for separating certain 'reactive' amino-acids, for example, leucine and glycine. On adding dilute acetic acid to an alkaline solution of the two derivatives, 3:5-dinitrobenzoylleucine was precipitated while 3:5-dinitrobenzoylglycine, which remained in solution, could be precipitated by dilute HCl.

The principle of fractional precipitation at different *pH* has recently been extended by Town [1941] to the separation of the isomers of valine and leucine. In this paper, Town claims that, in the presence of NaOH, 'reaction times for the benzoylation of the order of 30 min. have been found'. This is contrary to our experience, which has shown that one

great advantage of this reagent as compared with most other similar reagents, is that the reaction is extremely rapid and is usually complete in less than 2 min.

It is difficult to see how, in the presence of NaOH, the 'reaction time' can be greater than 2 min., for we have shown that 1 mol. of 3:5-dinitrobenzoyl chloride, in the absence of amino-acid, is almost completely dissolved and hydrolysed by 2 mol. NaOH in 2 min. Hence, if in the 3:5-dinitrobenzoylation of an amino-acid, the latter does not react within 2 min., the acid chloride uses up the alkali by the process of rapid hydrolysis, and no further reaction can take place.

It should be pointed out that our technique differs from that of Town in that we add the powdered acid chloride in one lot to the alkaline solution of the amino-acid. Town, on the other hand, adds the acid chloride in small quantities at a time, but this in itself cannot cause the actual 'reaction time' to be longer.

Our experiments show that with a solution of glycine in NaOH, no advantage is gained by adding the acid chloride in small quantities at a time over 30 min.

One mol. of acid chloride requires 2 mol. NaOH for hydrolysis, and so it seemed of interest to compare the amounts of certain acid chlorides (used in amino-acid identification) left after 2 min. when acted upon by a slight excess of *N* NaOH. From the results given in the Experimental Section, it will be seen that among the aromatic acid chlorides, 3:5-dinitrobenzoyl chloride and *m*- and *p*-nitrobenzoyl chlorides are in a class by themselves, the hydrolysis being complete within this period. Of course, it is not to be assumed that the reaction of a particular acid chloride with the OH<sup>-</sup> ion will exactly correspond to that with an —NH<sub>2</sub> group, as far as order of reactivity is concerned.

The reaction between 3:5-dinitrobenzoyl chloride and aniline in ethyl acetate solution, and also that between benzoyl chloride and aniline, in the same solvent, have been investigated. Ethyl acetate was chosen as solvent because of the high solubility of the anilides produced and the almost complete insolubility of aniline hydrochloride in this liquid. The yields of aniline hydrochloride which had separated after 2 min with 3:5-dinitrobenzoyl chloride and benzoyl chloride were 95 and 78 % of the possible amount, respectively. Under similar conditions, using *p*-toluenesulphonyl chloride, no aniline hydrochloride had separated after 2 min. Even with methanesulphonyl chloride (which is very reactive toward OH<sup>-</sup> ions) no precipitate appeared within 2 min. It is proposed, at a later stage, to investigate this phenomenon in greater detail.

The 3:5-dinitrobenzoylation of aniline was also examined. Aniline was chosen because of the ready separation of 3:5-dinitrobenzanilide, in a reasonably pure condition, from the reaction mixture, which contained varying quantities of aqueous NaOH. It was again shown that no advantage was to be gained by adding the acid chloride in small quantities over 30 min.

It was pointed out in our previous communications that very little 3:5-dinitrobenzoic acid was formed during the acylation of the more reactive amino-acids. On the other hand, with the less reactive amino-acids, considerable quantities of 3:5-dinitrobenzoic acid were sometimes produced. In support of this, the yields of 3:5-dinitrobenzoic acid obtained, were frequently quoted, and the fact that derivatives precipitated by dilute acetic acid were free from 3:5-dinitrobenzoic acid, was stressed.

In looking for a possible alternative rapid reagent for the identification of amino-acids and one which would give a product free from organic acid on acidifying with HCl, it seemed that 3:5-dinitrobenzenesulphonyl chloride might meet the case, as the corresponding acid is very soluble in water. Its reactivity, however, was found to be of a low order.

Methanesulphonyl chloride was found to be very reactive towards OH<sup>-</sup> ions. The possibility of using this reagent for identifying amino-acids was first investigated by Helferich & Mittag [1938], but a quicker method for obtaining the glycine derivative (although in smaller yield) than that described by these authors, is given below. However,

one drawback to the general use of this reagent in the case of amino-acids, is the high solubility of the methanesulphonyl derivatives in water. Furthermore, from work so far done, it appears that methanesulphonyl chloride is less reactive towards the  $\text{NH}_2$  group than towards  $\text{OH}^-$  ions. It seems that methanesulphonyl chloride is a suitable and very rapid reagent for the identification of certain phenols. It does not appear to react to any extent with salicylic acid or with tyrosine, in which respect it resembles 3:5-dinitrobenzoyl chloride.\* In this connexion, it should be noticed that with substances (e.g. phenol, aniline) which give 3:5-dinitrobenzoyl derivatives insoluble in  $\text{NaOH}$ , no reaction of any appreciable extent takes place if the reactants are shaken together in a stoppered bottle in the usual way. The solid acid chloride becomes coated immediately with a thin layer of the derivative, thus preventing any further reaction. The reaction can be caused to take place if the reactants are thoroughly ground together in a mortar.

The higher aliphatic sulphonyl chlorides, such as ethane-, *n*-propane- and *n*-butanesulphonyl chlorides, give derivatives of phenols of lower m.p. and, moreover, the *n*-butanesulphonyl chloride shows a marked decrease in reactivity.

$\alpha$ -Toluenesulphonyl chloride is slightly less reactive than methanesulphonyl chloride, but the reaction with glycine is complicated by the simultaneous production of sodium  $\alpha$ -toluenesulphonate.

Town's paper [1941] is also concerned with the isolation from a specimen of *dl*-valine of a substance which he believed to be 3:5-dinitrobenzoyl*nor*valine, and from a specimen of leucine, of a substance which he suggested might be 3:5-dinitrobenzoyl*nor*leucine, the identity of both these compounds being subject to confirmation. We have prepared the 3:5-dinitrobenzoyl derivatives of *dl*-*nor*valine (*dl*- $\alpha$ -amino-*n*-valeric acid) and of *dl*-*nor*leucine (*dl*- $\alpha$ -amino-*n*-hexoic acid) and we find that their melting points do not correspond with those recorded by Town. In this connexion, the possibility of dimorphism among dinitrobenzoyl derivatives must not be overlooked, particularly as dimorphism among other dinitro compounds is known, as, for example, the dimorphism of the 2:4-dinitrophenylhydrazone of acetaldehyde [Bryant, 1933; Martin *et al.* 1941].

The melting points of a few of the derivatives obtained by one of us [Saunders, 1934; 1938] are different from those recorded in the literature [Town, 1941]. 3:5-Dinitrobenzoyl derivatives have a marked tendency to form hydrates, and these differences of melting point may be due to the separation of different hydrates.

It should also be noted that the melting points of certain hydrates depend upon the rate of heating, as already reported, e.g. for 3:5-dinitrobenzoylsarcosine [Saunders, 1934].

## EXPERIMENTAL

### *3:5-Dinitrobenzoic acid*

Benzoic acid (50 g.) was dissolved in conc.  $\text{H}_2\text{SO}_4$  (230 ml.) in a litre flask which was then fitted with a ground-in condenser. Nitric acid (73 ml., sp.gr. 1.5) was then added, a few ml. at a time, the flask being well shaken and cooled in ice-water during the addition. Much heat was evolved and a clear yellow solution was obtained. After adding porous pot, the mixture was heated gradually in a water bath up to  $100^\circ$  during 45 min. At  $70$ – $80^\circ$  the reaction tended to become vigorous and was moderated, when necessary, by immersing the flask in cold water. The mixture was maintained at  $100^\circ$  for 15 min., with occasional shaking, and then transferred to an oil bath at  $100^\circ$  and the temperature raised to  $130^\circ$  over 30 min. and then kept between  $130$  and  $140^\circ$  for 1 hr. (The total period of heating was thus  $2\frac{1}{2}$  hr.) The flask was allowed to cool and at about  $90^\circ$ , crystals began to separate, the process of crystallization being accelerated by shaking. When cold, the

\* Unlike salicylic acid, methyl salicylate is readily dinitrobenzoylated. It has also now been shown that 3:5-dinitrobenzoyl chloride does not react with mandelic acid in the presence of  $\text{NaOH}$  to any appreciable extent.

contents of the flask were poured into 3–4 l. of ice-water and the crystals filtered off. After washing with water and drying they had m.p. 204° and were sufficiently pure, without recrystallization, for most purposes. (Found: C, 39.6; H, 2.04. Calc. for  $C_7H_4O_6N_2$ : C, 39.6; H, 1.90 %.) After recrystallization from water, the acid was obtained in characteristic rhombs. m.p. 204°.

### *3:5-Dinitrobenzoyl chloride*

3:5-Dinitrobenzoic acid (30 g., 1 equiv.) was mixed with  $PCl_5$  (33 g., 1.1 equiv.) and the mixture was heated under reflux in an oil bath at 120–130° for 1½ hr. The phosphorus oxychloride was then distilled off at 25° (20 mm.), the temperature of the bath being finally raised to 110° during this process. On cooling, the residue in the flask solidified to a brown mass of crude 3:5-dinitrobenzoyl chloride (yield, theoretical; m.p. 63° to a clear liquid). Carbon tetrachloride proved to be a very suitable solvent for recrystallization, particularly for large quantities. Yield of recrystallized material, 25 g. (77 %), m.p. 66–68°. Beautiful long white needles of pure material, with m.p. 69.5°, were obtained by recrystallization from light petroleum (b.p. 40–60°), but a relatively large amount of solvent was required. (Found: N, 12.2; Cl, 15.5. Calc. for  $C_7H_3O_5N_2Cl$ : N, 12.2; Cl, 15.4 %.)

### *3:5-Dinitrobenzoyl dl- $\alpha$ -amino-n-valeric acid*

The amino-acid (1.17 g., 0.01 mol.) was dissolved in *N* NaOH (20 ml., 0.02 mol.) and finely powdered 3:5-dinitrobenzoyl chloride (2.31 g., 0.01 mol.) added. The mixture was shaken in a stoppered bottle for 2 min. Practically the whole of the solid dissolved very rapidly, and after filtration, the solution was acidified with dilute acetic acid. The white precipitate (2.12 g.) thus obtained was twice recrystallized from aqueous methyl alcohol. m.p. 227.5–228.5° to a brownish liquid. (Found: C, 46.5; H, 4.38.  $C_{12}H_{13}O_7N_3$  requires: C, 46.3; H, 4.22 %.)

### *3:5-Dinitrobenzoyl dl- $\alpha$ -amino-n-hexoic acid*

The derivative was prepared similarly to that of *dl- $\alpha$ -amino-n-valeric acid*. Yield of crude compound, 77.3 %. Recrystallized twice from aqueous ethyl alcohol. m.p. 203.5–204°. (Found: C, 47.9; H, 4.92.  $C_{13}H_{15}O_7N_3$  requires: C, 48.0; H, 4.65 %.)

### *3:5-Dinitrobenzoyl dl- $\alpha$ -methyl- $\alpha$ -amino-n-butyric acid*

Prepared from *dl- $\alpha$ -methyl- $\alpha$ -amino-n-butyric acid* (0.88 g., 0.0075 mol.) dissolved in *N* NaOH solution (15 ml.) and mixed with 3:5-dinitrobenzoyl chloride (1.73 g., 0.0075 mol.). On acidifying the alkaline solution with dilute acetic acid, a white precipitate (A, 0.91 g.) was obtained. On further acidification of the filtrate from A with dilute hydrochloric acid, 3:5-dinitrobenzoic acid was obtained, the yield being 0.7 g. Two recrystallizations of A from aqueous methyl alcohol gave white flakes. m.p. 186°. (Found: C, 46.4; H, 4.53.  $C_{12}H_{13}O_7N_3$  requires: C, 46.3; H, 4.22 %.)

### *3:5-Dinitrobenzoylglycine*

(i) Prepared according to our previous method [1934] by shaking glycine (0.38 g.) in *N* NaOH solution (10 ml.) with 3:5-dinitrobenzoyl chloride (1.16 g.) for 2 min. Very little acid chloride remained to be filtered off. Yield of product on acidification: 1.17 g. (86 %). m.p. of crude material: 169–175°.

We originally gave the melting point of the recrystallized material as 179° which is close to Adamson's [1939] figure of 178°. Town gives m.p. 182.4°. We now find that even after three recrystallizations (10 % acetic acid, water, water plus animal charcoal) the melting point is substantially the same at 179.5°.

(ii) Prepared as above but the acid chloride was added in small quantities at a time over 30 min. Yield of product: 1.14 g. (84 %). m.p. 168–175°.

*Reaction between various acyl chlorides and N sodium hydroxide solution*

To a weighed quantity of the acyl chloride, a measured volume of *N* NaOH solution was added and the mixture shaken for 2 min. The unchanged chloride (if solid) was then immediately filtered off, washed, dried and weighed.

The solids were recrystallized to constant melting point before the experiment and the melting point of the unreacted material was taken in each case to ensure that it had undergone no essential change. The solid chlorides were finely powdered before use. In the experiments with liquid chlorides, the unchanged substances were extracted after 2 min. by quickly pouring the mixture into 25 ml. of ether in a separating funnel and shaking. After separating and drying over calcium chloride, the ether was distilled off from a previously weighed flask which was then gently reheated and allowed to cool in a dry atmosphere until a constant weight was obtained.

The results, which are, of course, only roughly quantitative, are given in the accompanying table.

| Acid chloride used            | Amount of chloride used<br>g. | Amount of <i>N</i> NaOH<br>ml. | Amount of chloride left<br>after 2 min.<br>g. | Percentage which had reacted |
|-------------------------------|-------------------------------|--------------------------------|---|------------------------------|
| Methanesulphonyl              | 1.15                          | 25                             | Disappeared almost immediately                | 100                          |
| Ethanesulphonyl               | 1.29                          | 25                             | Disappeared almost immediately                | 100                          |
| <i>n</i> -Propanesulphonyl    | 1.43                          | 25                             | Disappeared within 30 sec.                    | 100                          |
| $\alpha$ -Toluenesulphonyl    | 0.95                          | 12.5                           | 0.06  | 94.2                         |
| $\beta$ -Naphthalenesulphonyl | 1.13                          | 12.5                           | 1.04  | 8.0                          |
| <i>p</i> -Toluenesulphonyl    | 0.95                          | 12.5                           | 0.88  | 7.4                          |
| 3:5-Dinitrobenzenesulphonyl   | 0.67                          | 6.25                           | 0.58  | 13.4                         |
| Benzoyl                       | 1.41                          | 25                             | 0.44  | 68.8                         |
| <i>o</i> -Nitrobenzoyl        | 1.86                          | 25                             | 0.21  | 88.7                         |
| <i>m</i> -Nitrobenzoyl        | 0.93                          | 12.5                           | 0.00  | 100                          |
| <i>p</i> -Nitrobenzoyl        | 0.93                          | 12.5                           | Disappeared within 90 sec.                    | 100                          |
| 3:5-Dinitrobenzoyl            | 1.16                          | 12.5                           | Disappeared in about 40 sec.                  | 100                          |
| 3:5-Dinitrobenzoyl            | 1.16                          | 10                             | 0.01  | 99.1                         |

*Reaction between (i) 3:5-dinitrobenzoyl chloride and aniline in ethyl acetate solution,  
(ii) benzoyl chloride and aniline in ethyl acetate solution*

The reaction considered was:  $2C_6H_5NH_2 + RCOCl = C_6H_5NH_2 \cdot HCl + RCONHC_6H_5$ .

Aniline (0.93 g., 0.01 mol.) and 3:5-dinitrobenzoyl chloride (1.15 g., 0.005 mol.) were each dissolved in separate quantities of 40 ml. of ethyl acetate. On mixing the solutions, a white precipitate of aniline hydrochloride came down immediately. After 2 min., this was filtered off as rapidly as possible and, after drying, had m.p. 197–198° (m.p. of aniline hydrochloride, 198°). Light petroleum (b.p. 60–80°) added to the filtrate produced a white crystalline precipitate of m.p. 234° (3:5-dinitrobenzanilide has m.p. 234°).

An exactly parallel experiment to the above, but using the correct molar quantity of freshly distilled benzoyl chloride (b.p. 197.5–198°) in place of the 3:5-dinitrobenzoyl chloride, was carried out.

The results obtained in each case are given below:

| Amount of aniline<br>in 40 ml. of ethyl<br>acetate<br>g. | Amount of 3:5-<br>dinitrobenzoyl<br>chloride in 40 ml.<br>of ethyl acetate<br>g. | Amount of benzoyl<br>chloride in 40 ml.<br>of ethyl acetate<br>g. | Yield of<br>aniline HCl<br>% | m.p. of anilide |
|--|--|---|------------------------------|-----------------|
| 0.93   | 1.15   | —   | 95                           | 234°            |
| 0.93   | —  | 0.70  | 80                           | 162°            |

*Reaction between aniline and 3:5-dinitrobenzoyl chloride*

In Exp. I, aniline, 3:5-dinitrobenzoyl chloride and *N* NaOH were ground together in a mortar for 2 min. and the precipitated 3:5-dinitrobenzanilide was filtered off, washed well, and again ground with 20 ml. of *N* NaOH to remove any unchanged acid chloride,

filtered off, and the two filtrates combined. This solution was concentrated and acidified with dilute HCl to precipitate the 3:5-dinitrobenzoic acid formed by hydrolysis of the acid chloride. In Exps. II and III, the NaOH was in excess and so it was not necessary to grind up the precipitated 3:5-dinitrobenzanilide with an extra amount of NaOH in order to remove unchanged acid chloride. Exp. IV resembled Exp. I except that the acid chloride was added in small quantities at a time over 30 min.

| Exp. | Amount of acid | Amount of     | Yield of 3:5-      | Yield of 3:5-       |
|------|----------------|---------------|--------------------|---------------------|
|      | chloride       | <i>N</i> NaOH | dinitrobenzanilide | dinitrobenzoic acid |
|      | g.             | ml.           | %                  | %                   |
| I    | 0.93           | 10            | 72                 | 21                  |
| II   | 0.93           | 20            | 70                 | 20                  |
| III  | 0.93           | 30            | 66                 | 24                  |
| IV   | 0.93           | 10            | 68                 | 23                  |

#### *3:5-Dinitrobenzoyl methyl salicylate*

Methyl salicylate (1.52 g., 0.01 mol.), 3:5-dinitrobenzoyl chloride (2.31 g.) and *N* NaOH (10 ml.) were ground together in a small mortar for 2 min. The reaction mixture, an orange liquid and a paste-like mass, was treated with ether and the white solid so obtained was filtered off and twice recrystallized from a mixture of chloroform and light petroleum (B.P. 40–60°). It formed small, colourless needles; m.p. 107.5°. (Found: C, 52.0; H, 3.04.  $C_{15}H_{10}O_8N_2$  requires C, 52.0; H, 2.91 %.)

#### *Reaction between 3:5-dinitrobenzoyl chloride and dl-mandelic acid*

*dl*-Mandelic acid (1.52 g., 0.01 mol.) was dissolved in *N* NaOH (30 ml., 0.03 mol.) and powdered 3:5-dinitrobenzoyl chloride (2.31 g., 0.01 mol.) was added and the mixture shaken for 2 min. A small quantity (0.13 g.) of unchanged acid chloride was filtered off and the filtrate acidified with dilute HCl. After cooling and standing for some time, a pale yellow precipitate, A, separated. The filtrate from A was evaporated to dryness and the solid so obtained extracted with boiling acetone. The part (NaCl, 1.65 g.) insoluble in acetone was filtered off and the solution was evaporated to dryness, producing a slightly sticky solid which was recrystallized from benzene in white flakes (B).

Substance A (1.91 g.) had m.p. 204–205°, and proved to be 3:5-dinitrobenzoic acid. Yield: 95.7%, allowing for the small quantity of acid chloride recovered unchanged. Substance B (1.33 g.) had m.p. 104–114°, and when once recrystallized from benzene, melted at 118–119° (*dl*-mandelic acid has m.p. 118°). Yield 87.5%.

#### *m-Nitrobenzoylglycine*

This substance was prepared by direct *m*-nitrobenzoylation, a method which does not seem to have been used previously. It had been prepared from benzoylglycine by nitration [Conrad, 1877], and also from aminoacetonitrile and *m*-nitrobenzoyl chloride, followed by hydrolysis [Klages & Haack, 1903].

Glycine (0.75 g.) was dissolved in *N* NaOH (20 ml.) and powdered *m*-nitrobenzoyl chloride (1.86 g., 0.01 mol.) added. The whole was shaken for 2 min., after which practically all the acid chloride had dissolved. The filtered liquid gave no precipitate with acetic acid, but on the addition of dilute HCl, an emulsion formed and soon deposited white crystals (1.86 g.). These had m.p. 161–163°. After two recrystallizations from water, white needles were obtained having m.p. 166°. Melting points given in the literature are: 162° [Conrad], 165° [Klages & Haack], 165–167° [Sieber & Smirnow, 1887].

#### *3:5-Dinitrobenzenesulphonyl glycine*

The 3:5-dinitrobenzenesulphonic acid was prepared by the method of Griffith [1924], and converted into the sulphonyl chloride by the method of Jackson & Earle [1903].

Glycine (0.75 g., 0.01 mol.) was dissolved in *N* NaOH (30 ml., 0.03 mol.) and 3:5-dinitrobenzenesulphonyl chloride (2.67 g., 0.01 mol.) was added, the mixture being well shaken for 1½ hr. The 3:5-dinitrobenzenesulphonyl chloride (0.06 g., m.p. 103°) which had not dissolved, was filtered off and the filtrate acidified with dilute HCl, being then evaporated on the water bath until crystallization commenced. The brownish crystals which separated were extracted with ether, leaving a residue (2.4 g.). This residue was recrystallized, in small yellow flakes, from a relatively large volume of alcohol. (Found: N, 10.1. Sodium 3:5-dinitrobenzenesulphonate, C<sub>6</sub>H<sub>3</sub>O<sub>7</sub>N<sub>2</sub>SNa, requires: N, 10.4 %.)

The ethereal extract, on evaporation to dryness, gave a substance (0.25 g.) which was recrystallized once from ether and twice from water to give almost colourless flakes with m.p. 191–192°. (Found: C, 32.0; H, 2.38; N, 14.1. The glycine derivative, C<sub>8</sub>H<sub>7</sub>O<sub>8</sub>N<sub>3</sub>S, requires: C, 31.5; H, 2.32; N, 13.8 %.)

#### *Methanesulphonyl glycine*

The methanesulphonyl chloride was prepared from methyl thiocyanate [Johnson & Douglass, 1939].

Glycine (0.75 g., 0.01 mol.) was dissolved in *N* NaOH solution (20 ml., 0.02 mol.) and methanesulphonyl chloride (1.15 g., 0.01 mol.) added, and the whole shaken for 2 min. A clear, colourless solution was produced and acidified with dilute HCl, but as no precipitate appeared, even on standing, the liquid was evaporated on the water bath until crystals formed on cooling. After drying, they were extracted with boiling alcohol to separate any sodium chloride which they might have contained. From the filtered extract, white platelets separated and were halogen-free. After a second recrystallization from alcohol, the compound had m.p. 172–173°. (Found: C, 23.8; H, 4.75. Calc. for C<sub>8</sub>H<sub>7</sub>O<sub>4</sub>NS: C, 23.5; H, 4.61 %.)

#### *Methanesulphonyl anthranilic acid*

Prepared similarly to the glycine derivative. After 2 min. shaking, the clear solution was acidified with dilute HCl, when a whitish, granular precipitate separated. It was recrystallized from water (charcoal) in radiating tufts of white needles: m.p. 190.5–191.5°. (Found: C, 44.5; H, 4.40. C<sub>8</sub>H<sub>9</sub>O<sub>4</sub>NS requires: C, 44.6; H, 4.22 %.)

#### *Phenyl methanesulphonate*

Phenol (1.88 g., 0.02 mol.) was dissolved in *N* NaOH (30 ml., 0.03 mol.) and methanesulphonyl chloride (2.29 g., 0.02 mol.) was added. A turbidity was immediately produced and a white crystalline precipitate (2.01 g.) separated during 2 min. shaking. In the process of recrystallization from water, the substance tended to appear first as an oil which gave small white flaky crystals on further cooling: m.p. 59.5°. (Found: C, 48.9; H, 5.10. C<sub>7</sub>H<sub>8</sub>O<sub>3</sub>S requires: C, 48.8; H, 4.68 %.)

#### *p-Nitrophenyl methanesulphonate*

Prepared similarly to the phenol derivative, from *p*-nitrophenol (1.39 g., 0.01 mol.) suspended in *N* NaOH (15 ml., 0.015 mol.), and methanesulphonyl chloride (1.15 g., 0.015 mol.). The crude product was washed well with water and recrystallized from 90 % ethyl alcohol in practically colourless crystals: m.p. 93–93.5°. They contained N and S, and gave no coloration with ferric chloride solution. (Found: C, 38.7; H, 3.32. C<sub>7</sub>H<sub>7</sub>O<sub>5</sub>NS requires: C, 38.8; H, 3.25 %.)

#### *α-Toluenesulphonyl glycine*

The sulphonyl chloride was prepared according to the method of Johnson & Ambler [1914].

Glycine (0.75 g.) was dissolved in *N* NaOH (30 ml., 0.03 mol.) and shaken with α-toluenesulphonyl chloride (1.91 g., 0.01 mol.). After shaking for 15 min., the solution

was filtered from a small quantity of unchanged chloride, acidified with dilute HCl and evaporated until crystals separated. These were extracted with boiling chloroform. The undissolved portion (1.1 g.) was recrystallized from methyl alcohol as white flaky crystals. They proved to be sodium  $\alpha$ -toluenesulphonate. (Found: C, 43.1; H, 3.85.  $C_7H_7O_3SNa$  requires: C, 43.3; H, 3.64 %.)

The chloroform solution deposited crystals (0.18 g.) on concentration. These were twice recrystallized from chloroform and had m.p. 152°. The substance was  $\alpha$ -toluenesulphonylglycine. (Found: C, 47.3; H, 5.09.  $C_9H_{11}O_4NS$  requires: C, 47.2; H, 4.84 %.)

## SUMMARY

1. An improved method for preparing 3:5-dinitrobenzoic acid, involving a great reduction in the time of nitration, is described.

2. The preparation of 3:5-dinitrobenzoyl chloride is described.

3. Our previous observation [Saunders, 1934; 1938] that the 3:5-dinitrobenzoylation of amino-acids in normal sodium hydroxide solution is complete in 2 min., has been confirmed. 3:5-Dinitrobenzoyl chloride itself is completely dissolved and hydrolysed by a slight excess of normal sodium hydroxide solution within 2 min.

4. The 3:5-dinitrobenzoyl derivatives of *dl*- $\alpha$ -amino-*n*-valeric, *dl*- $\alpha$ -amino-*n*-hexoic and *dl*- $\alpha$ -methyl- $\alpha$ -amino-*n*-butyric acids are described. The two first-named derivatives do not possess the melting points of the compounds isolated by Town [1941] from specimens of valine and leucine, and described by him as probably being 3:5-dinitrobenzoyl*nor*valine and 3:5-dinitrobenzoyl*nor*leucine respectively.

5. There is evidence to show that the 3:5-dinitrobenzoyl derivatives of certain amino-acids can exist in different forms, particularly where hydrates are concerned.

6. The relative merits of 3:5-dinitrobenzenesulphonyl chloride,  $\alpha$ -toluenesulphonyl chloride, methanesulphonyl chloride and other acid chlorides as acylating agents for amino-acids are discussed.

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